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(21) International Application Number: PCT/GB92/02252 (22) International Filing Date: 4 December 1992 (04.12.92) (30) Priority data: 9125768.3 4 December 1991 (04.12.91) GB (71)(72) Applicants and Inventors: HALE, Geoffrey [GB/GB]; WALDMANN, Herman [GB/GB]; Cambridge University, Department of Pathology, Immunology Division, Tennis Court Road, Cambridge CB2 1QP (GB). (74) Agent: MARCHANT, James, Ian; Elkington and Fife, Prospect House, 8 Pembroke Road, Sevenoaks, Kent TN13 1XR (GB).		(81) Designated States: AU, CA, JP, KR, US, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE). Published <i>With international search report.</i>
(54) Title: CDw52-SPECIFIC ANTIBODY FOR TREATMENT OF MULTIPLE SCLEROSIS (57) Abstract The invention relates to the use of an antibody recognising the CDw52 antigen in the treatment of multiple sclerosis. Preferably the antibody is the humanised antibody in the humanised antibody CAMPATH-1H.		

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CDw52 - SPECIFIC ANTIBODY FOR TREATMENT OF MULTIPLE SCLEROSIS

The present invention relates to a method of treatment of certain auto-immune conditions in a human or animal subject.

5 The cells and molecules in an animal body do not normally stimulate an adaptive immune response in that body as a result of a number of mechanisms which ensure a state referred to as "self-tolerance". However, in certain
10 circumstances these mechanisms may fail to prevent the stimulation of such an immune response and this condition is referred to as "auto-immunity". A number of diseases in humans and animals are now thought to result from auto-immunity.

15 Multiple sclerosis is a demyelinating disease of the central nervous system the onset of which generally occurs within the age range from about 15 to 45. Myelin is a fatty substance which forms a sheath around certain nerve fibres and which conducts nervous impulses at a rate which enables muscles to make precise and delicate movements.
20 The disease is characterised by induration of the sheath substance leading to the formation of plaques of varying size and location which interfere with the impulses normally conducted by the sheath. The course of the disease can be highly variable in individual patients and may be subject to periods of spontaneous remission.
25 However, the disease generally results in progressive deterioration of the control of muscle function with ultimate paralysis in many cases. The precise cause of multiple sclerosis is unknown and it may result from a complex interaction of a number of different factors.
30 However, it is now generally recognised that there is at least an autoimmune component in the causation of the disease.

35 The CDw52 antigen (a 23kDa glycoprotein also referred to as CAMPATH-1) is strongly expressed on the surface of all human lymphocytes and most monocytes but is absent from

other blood cells including stem cells. A number of antibodies have been developed directed against CDw52 which is an unusually good target for complement-mediated attack. Antibodies against CDw52 bind to all lymphocytes and monocytes but lyse only lymphocytes (T and B) in vivo. Antigens similar to CDw52 are expressed in other mammalian species.

Antibodies which have been developed against CDw52 and which have been used in therapy include the following:

CAMPATH-1M is a rat IgM monoclonal antibody which has been used extensively in vitro for purging bone marrow harvests in order to deplete the T cell population prior to bone marrow transplantation. Marked reduction in the incidence and severity of graft-versus-host disease has been seen with this therapy.

CAMPATH-1G is a rat IgG2b class-switch variant of an IgG2a antibody recognising the CDw52 antigen which has been used in vivo to achieve immunosuppression in more than 100 patients undergoing organ and bone marrow transplantation, management of organ rejection and treatment of haematologic malignancies with a high level of success. However, the rapid development of an anti-rat immunoglobulin response, including the possibility of anaphylaxis, is likely to limit the use of rat monoclonal antibodies against the CDw52 antigen in humans in vivo.

CAMPATH-1H is a genetically manipulated IgG antibody obtained by grafting the complementarity determining regions from CAMPATH-1G into human framework regions. The resulting "humanized" antibody is highly effective in vitro being equivalent to the rat monoclonal antibody at complement lysis and two to four times better in cell-mediated lysis of human lymphocytes. No limiting anti-globulin response is anticipated with this humanized antibody.

Expression of CAMPATH-1H was achieved initially in rat myeloma cells by placing DNA encoding the engineered

antibody chains in genomic context under control of the immunoglobulin promoter/enhancer. CAMPATH-1H has recently been expressed to high levels in Chinese hamster ovary (CHO) cells.

5 It has now surprisingly been found that antibodies against the CDw52 antigen may be effective in the treatment of multiple sclerosis.

10 The present invention provides a method of treatment of a human subject suffering from multiple sclerosis which comprises administering to the said subject an effective amount of an antibody recognising the CDw52 antigen.

 The present invention also provides the use of an antibody recognising the CDw52 antigen for the manufacture of a medicament for the treatment of multiple sclerosis.

15 It is preferred to use an anti-CDw52 antibody which does not carry with it the risk of the development of an immune reaction against the anti-body itself. Accordingly whilst it is possible to use mouse or rat mono-clonal antibodies it is preferred to use antibodies which have
20 been produced by recombinant DNA technology and which have been engineered to reduce the risk of causing an immune reaction. Thus it is possible to use a chimeric antibody in which the constant domains of a mouse or rat anti-CDw52 antibody have been replaced by the constant domains of a
25 human antibody. However, it is preferred to use a humanized or CDR-grafted antibody for the treatment of humans, i.e. an antibody in which the complementarity determining regions from a mouse or rat antibody are combined with framework regions and constant domains from
30 one or more human antibodies.

 Most preferably the method according to the invention is carried out using the CDR-grafted antibody CAMPATH-1H (see Riechmann et al, Nature, 322, 323-327 (1988)). As noted above, the antibody CAMPATH-1H can be produced in rat
35 myeloma cells as originally described or it can be produced in any other expression system, particularly an expression

system suitable for the production of a correctly folded, glycosylated, mammalian protein. High yields of CAMPATH-1H have been obtained by expression in a genetically manipulated CHO cell line (Page and Sydenham, Biotechnology, 9, 64-68 (1991)).

The dosages of anti-CDw52 antibody, preferably CAMPATH-1H, to be administered to a patient suffering from multiple sclerosis will vary with the precise nature of the condition being treated and the recipient of the treatment. The dose will generally be in the range 1 to about 100mg for an adult patient, usually administered daily for a period between 1 and 30 days. The preferred daily dose is 1 to 10mg per day although in some instances larger doses of up to 40mg per day may be used.

The anti-CDw52 antibody, particularly CAMPATH-1H will generally be administered to the patient in the form of a pharmaceutical formulation. Such formulations preferably include, in addition to the antibody, a physiologically acceptable carrier or diluent, possibly in admixture with one or more other agents such as other antibodies or drugs, such as an antibiotic. Suitable carriers include, but are not limited to, physiological saline, phosphate buffered saline, phosphate buffered saline glucose and buffered saline. Alternatively the antibody may be lyophilised (freeze dried) and reconstituted for use when needed by the addition of an aqueous buffered solution as described above. Routes of administration are routinely parenteral, including intravenous, intramuscular, subcutaneous and intraperitoneal injection or delivery.

The anti-CDw52 antibody may be administered in combination with or sequentially with other drugs, in particular other drugs conventionally used in the treatment of multiple sclerosis, such as steroids, for example hydrocortisones such as methyl prednisolone, or other medicaments for the alleviation of specific symptoms of the disease. Sequential administration of the antibody with

another drug may be appropriate in some cases.

The invention is illustrated by the following case history of a patient treated with the CDR-grafted anti-CDW52 antibody CAMPATH-1H. The antibody used was produced by expression in a recombinant CHO cell line (Page and Sydenham, Biotechnology, 9, 64-68 (1991)) followed by extraction from the cell culture medium and purification.

It is to be understood that the following case history is not intended to be in any way limiting on the scope of the invention.

CASE HISTORY

The patient was a 43 year old female who sought medical attention after 10 days of progressive loss of sensation in her perineum. She had also experienced several episodes of urinary and fecal incontinence. Clinical evaluations were remarkable for normal visual evoked responses, multiple white matter abnormalities in the cerebral hemispheres identified by brain MRI, and oligoclonal bands in the CSF; a diagnosis of multiple sclerosis was made.

Treatment with high-dose iv methylprednisolone induced an incomplete recovery (decreased incontinence with continued perineal numbness and stiff legs). Four months after initially seeking medical attention, the patient had an episode of left-sided facial pain and left leg numbness consistent with brainstem demyelination. Over the next twelve months symptoms gradually worsened and at the end of this period the patient was admitted to hospital with left optic neuritis and Lhermitte's phenomenon. A second course of iv methylprednisolone (500 mg/day for five days) was associated with limited improvement. The disease was judged to have entered a chronic progressive phase and over the next few months the patient suffered further identifiable episodes of brainstem demyelination and left optic neuritis.

Twenty months after she initially sought medical

attention, the patient received treatment with CAMPATH-1H (2 mg/day iv for five days, two days rest, then 10 mg/day iv for five days). Lymphocyte depletion was rapid, falling to 0.2×10^9 cells/L on the second dosing day from a pretreatment value of 3.8×10^9 cells/L. Lymphocyte counts remained less than 0.3×10^9 for approximately one month and then rose slowly to a value of 1.07×10^9 cells/L three months after treatment. CRP, within normal limits prior to treatment, increased to 140 mg/L, fell, and increased again to 91 mg/L on dosing days three and nine, respectively. This "double-peaked" pattern was also seen with neutrophils which increased during the same time course and coincided with the second and third dosing days at 2 and 10 mg respectively (10 mg doses followed a two day rest).

Disease activity was not significantly altered during or immediately following CAMPATH-1H treatment. Kurtzke neurological status at approximately one and two months after initiation of treatment was improved to a composite score of 11 from a pretreatment score of 14. A second rating system for scoring disease activity in demyelinating disease also documented improvement in the progression, disability and relapse assessment categories.

The 10 dose CAMPATH-1H treatment course was well tolerated. Fever and headache were reported as adverse events with both occurring with administration of the first 2 mg and 10 mg doses. Also reported was transient left-sided visual disturbance on day three, the significance of which was unclear in the light of the pre-existing left optic neuritis.

Approximately 18 months after the treatment with CAMPATH-1H, the improvement in the condition of the patient had been maintained.

CLAIMS:

1. The use of an antibody recognising the CDw52 antigen
5 for the manufacture of a medicament for the treatment of multiple sclerosis.
2. The use according to Claim 1 wherein the antibody is
10 a humanised antibody, including CDRs from a mouse or rat antibody combined with framework regions and constant domains from one or more human antibodies.
3. The use according to Claim 2 wherein the antibody is
15 CAMPATH-1H.
4. The use according to any of Claims 1 to 3 wherein the
treatment involves daily administration to a patient of a
dose of from 1 to 10 mg per day for a period of 1 to 30
20 days.
5. The use according to any of Claims 1 to 4 wherein the
treatment includes administration of a steroid.
6. A therapeutic agent for the treatment of multiple
25 sclerosis comprising an antibody recognising the CDw52 antigen as the active ingredient.
7. A therapeutic agent according to Claim 6 wherein the
antibody is a humanised antibody including CDRs from a
30 mouse or rat antibody combined with framework regions and constant domains from one or more human antibodies.
8. A therapeutic agent according to Claim 7 wherein the
antibody is CAMPATH-1H.
35
9. A therapeutic agent according to any of Claims 6 to

8 wherein the daily dose of the antibody is from 1 to 10 mg per day for a period of 1 to 30 days.

5 10. A therapeutic agent as claimed in any of Claims 6 to 9 wherein the treatment includes administration of a steroid.

10 11. A method for the treatment of a human subject suffering from multiple sclerosis which comprises administering to the said subject an effective amount of an antibody recognising the CDw52 antigen.

15 12. A method according to Claim 11 wherein the antibody is a humanised antibody including CDRs from mouse or rat antibody combined with framework regions and constant domains from one or more human antibodies.

20 13. A method according to Claim 12 wherein the antibody is CAMPATH-1H.

25 14. A method according to any of Claims 11 to 13 which comprises daily administration of the antibody at a dose of from 1 to 10 mg per day for a period of 1 to 30 days.

15. A method according to any of Claims 11 to 14 which includes administration of an effective amount of a steroid.

I. CLASSIFICATION OF SUBJECT MATTER (If several classification symbols apply, indicate all) ⁶		
According to International Patent Classification (IPC) or to both National Classification and IPC Int.Cl. 5 A61K39/395; A61K31/57		
II. FIELDS SEARCHED		
Minimum Documentation Searched ⁷		
Classification System	Classification Symbols	
Int.Cl. 5	A61K ; C07K	
Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched ⁸		
III. DOCUMENTS CONSIDERED TO BE RELEVANT⁹		
Category ¹⁰	Citation of Document, ¹¹ with indication, where appropriate, of the relevant passages ¹²	Relevant to Claim No. ¹³
X	EP,A,0 328 404 (MEDICAL RESEARCH COUNCIL) 16 August 1989	1-3,6-9, 11-13
Y	see page 4, line 9 - line 14; claims ---	5,10,15
Y	JOURNAL OF NEUROIMMUNOLOGY vol. 22, no. 1, March 1989, AMSTERDAM, THE NETHERLANDS pages 1 - 9 W. WEBER ET AL. 'In vitro functional blocking of myelin basic protein-specific cytolytic human T lymphocyte clones by immunosuppressive drugs and monoclonal antibodies.' see the whole document --- -/--	5,10,15
<p>¹⁰ Special categories of cited documents:</p> <p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document but published on or after the international filing date</p> <p>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</p> <p>"O" document referring to an oral disclosure, use, exhibition or other means</p> <p>"P" document published prior to the international filing date but later than the priority date claimed</p> <p>"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</p> <p>"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step</p> <p>"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.</p> <p>"&" document member of the same patent family</p>		
IV. CERTIFICATION		
Date of the Actual Completion of the International Search 04 FEBRUARY 1993	Date of Mailing of this International Search Report 05.02.93	
International Searching Authority EUROPEAN PATENT OFFICE	Signature of Authorized Officer N00IJ F.J.M.	

III. DOCUMENTS CONSIDERED TO BE RELEVANT (CONTINUED FROM THE SECOND SHEET)

Category ^o	Citation of Document, with indication, where appropriate, of the relevant passages	Relevant to Claim No.
P,X	WO,A,9 207 084 (THE WELLCOME FOUNDATION LIMITED) 30 April 1992 see claims 1-7,15,16 see page 10 - page 11 -----	1-3,6-9, 11-13

INTERNATIONAL SEARCH REPORT

International application No.

PCT/GB92/02252

B x I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.: 4, 14
because they relate to subject matter not required to be searched by this Authority, namely:
see PCT-Rule 39.1(iv)
Remark: Although claims 11-13 and 15 are directed to a method of treatment of the human/animal body the search has been carried out and based on the alleged effects of the compound/composition.
2. ☐ Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO.

GB 9202252
SA 67614

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report.
The members are as contained in the European Patent Office EDP file on
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Patent document cited in search report	Publication date	Patent family member(s)	Publication date
EP-A-0328404	16-08-89	AU-A- 3062689	06-09-89
		WO-A- 8907452	24-08-89
		GB-A, B 2216126	04-10-89
WO-A-9207084	30-04-92	AU-A- 8729491	20-05-92
		EP-A- 0504363	23-09-92
		FR-A- 2668164	24-04-92
		FR-A- 2677997	24-12-92
		GB-A- 2253397	09-09-92